# DESCRIPTION OF THE THIRD INSTAR LARVA OF CERATITIS ROSA KARSCH (DIPTERA: TEPHRITIDAE)

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Abstract.—The mature third instar larva of *Ceratitis rosa* Karsch is described and illustrated. It is distinguished from the larva of the Mediterranean fruit fly, *Ceratitis capitata* (Wiedemann), by the presence of a minute subapical tooth on the mandible, the shape of the anterior and posterior spiracles, and the shape of the cephalic segment. Numbers of oral ridges and anterior spiracular tubules are similar for the two species.

*Key Words:* Natal fruit fly, Mediterranean fruit fly, immatures, diagnostic characters, scanning electron microscopy, morphology

The Natal fruit fly, Ceratitis rosa Karsch, is a major pest that is widespread in Africa, and has been introduced onto the islands of Mauritius and Reunion. It is polyphagous, and infests at least 27 of the same hosts infested by the Mediterranean fruit fly, Ceratitis capitata (Wiedemann) (White and Elson-Harris 1992). The only existing description of the immature stages of this species to date consists of a figure of the anterior spiracles (Orian and Moutia 1960). The mature third instar larva of C. rosa is here described and characters for distinguishing it from C. capitata are discussed, because of their economic importance, and the danger of misidentification.

### MATERIALS AND METHODS

Preserved mature third instar larvae of *C.* rosa were obtained from culture (see Specimens Examined). Series of specimens were prepared for study by dissecting, compound, and scanning electron microscopy. Slide mounted specimens (n = 7) were first slit along one side and around three-quarters of the caudal segment, macerated in

10% cold KOH, rinsed with water, passed through an alcohol series, and mounted from 95% alcohol into Euparal or Hoyer's medium. Specimens examined by SEM (n = 14) were rinsed in clean alcohol, pricked in the side with a minuten pin, passed through an alcohol series to absolute alcohol, then critical-point dried. Dried specimens were mounted on points of foil electrical tape, then carbon paint was applied to the connection between specimen and point to ensure good conductivity. Specimens were coated with gold or gold-palladium and examined with a JEOL-25 or AMRAY 1810 scanning electron microscope at 10 to 12.5 KeV.

Numbers of oral ridges and anterior spiracular tubules, dorsal spinule rows, and anal lobes were examined by dissecting microscope at a magnification of  $240\times$ , with the specimen (n = 30) removed from alcohol and dried until liquid had drained out of the oral ridges. Spinule rows were counted as the number of lines of spinules crossed by a longitudinal transect in the mid-dorsal region of the anterior margin of

the segment. Measurements for anterior and posterior spiracles were made on slidemounted specimens at a magnification of 250×. All measurements were calibrated with a stage micrometer. Specimens were compared with samples of C. capitata, ex culture from Programa Moscamed, Metapa, Chiapas, Mexico (n = 30, counts of oral ridges and anterior spiracular digits) and ex culture from the Tropical Fruit and Vegetable Research Laboratory, USDA, ARS, Honolulu, Hawaii (n = 10, SEM). Terminology for larval morphology follows Teskey (1981), Carroll and Wharton (1989) and White and Elson-Harris (1992, accessory plates). Voucher specimens are deposited at the National Museum of Natural History, Washington, D.C. (USNM) and Texas A&M University, College Station (TAMU).

### RESULTS

## Ceratitis rosa Karsch (Figs. 1–11)

## *Ceratitis rosa* Karsch 1887:22 *Pterandrus flavotibialis* Hering 1935:158

Diagnosis of third instar.—Medium-sized muscidiform larvae with minute subapical mandibular tooth; usually with 9–11 (rarely 8 or 12) oral ridges; accessory plates absent; leaf-like secondary stomal lobes present, sclerotized stomal guards absent; dorsal spinules present on segments T1-A1; anterior spiracles usually with 9–10 (rarely 7–8 or 11–12) tubules in a single straight row; base of anterior spiracle cylindrical, half as wide as apical width; posterior spiracles with rimae 2.75–3.8 times as wide as long; spiracular processes mostly unbranched; caudal ridge present; anal lobes entire.

Description of third instar.—Length 7.7– 9.6 mm; width 1.4–1.7 mm; creamy-white, subcylindrical, tapering gradually to cephalic segment; apparently amphipneustic (vestigial spiracular openings visible at high magnification on segments T3-A7 in slidemounted material).

*Head:* (Figs. 1, 7) with *cephalic lobes* moderately developed, in lateral view more

rounded and protuberant than in C. capitata (Fig. 7); antenna (Fig. 8) 2-segmented, both segments with sclerotized walls, the distal segment apically thin-walled and conical; maxillary palp (Fig. 8) with 2 large and 9 small sensilla visible in slide mounted material (visible by SEM as 3 papilla sensilla and 2 knob sensilla, the remainder as pits); dorsolateral group of sensilla with 2 papilla sensilla and a pit sensillum, adjacent to but distinct from palp; stomal organ (Fig. 2) with primary lobe small, bearing 3-4 unbranched peg sensilla, 1 campaniform sensillum, and 2 pit sensilla; 6 secondary lobes present: a broad, flat subtending lobe and a lobe medial to it; usually 2 additional lobes immediately surrounding the primary lobe anteriorly, and usually 2 lobes anteromedial to these, all with edges entire; none of these secondary lobes is strikingly similar to oral ridges; sclerotized stomal guards absent; labium short, triangular, with narrow lateral lobes; the median portion with 2 pairs of sensory organs; 6 pairs of other minute pit sensilla on the cephalic segment similar in arrangement to those of Anastrepha ludens (see Carroll & Wharton 1989), with one medial and two posterior to the cephalic lobes, and one each at the anterodorsal, posterodorsal, and posteroventral corners of the oral ridge region. Oral ridges usually 9-11 (but two had 8 and three had 12 ridges on one side only), well developed with margins scalloped to  $\frac{1}{4}-\frac{1}{5}$  of their depth (visible by SEM), located on a semicircular region laterad of mandible; accessory plates (supernumerary ridges) and other reticulation absent.

Cephalopharyngeal skeleton (Fig. 9) well developed; mandible black to dark brown, apical tooth pointed, with a small subapical tooth (visible in slide-mounted material); hypopharyngeal sclerite black in anterior half, bridge and posterior processes brown; tentoropharyngeal sclerite with dorsal and ventral cornua broadly joined, with strongly pigmented anterior and posterior margins, becoming less pigmented dorsally and ventrally; dorsal cornu split posteriorly;



Figs. 1–6. *Ceratitis rosa*, third instar. 1, Head (frontal view). 2, Stomal organ and associated lobes. 3, Caudal segment. 4, Anal lobes (caudal at top). 5, Anterior spiracles (anterolateral view). 6, Posterior spiracles (dorsal at top).



Figs. 7–8. Third instar. 7, Head (lateral view), a. *Ceratitis capitata*; b, *C. rosa* (ant, antenna; cl, cephalic lobe; md, mandible; mp, maxillary palp; so, stomal organ). 8, *C. rosa* (ant, antenna (lateral view); dlg, dorso-lateral group of sensilla; mp, maxillary palp (lateral and frontal views).

ventral cornu with a slight hump midway along dorsal margin, pigmented along dorsal margin and ventrally to slightly more than half its length, with an incomplete window; parastomal sclerite long, stout, brown, slightly hooked apically; other sclerites as follows: dental sclerite dark brown, narrow in profile, free from and distinctly posterior to base of mandible; labial sclerites dark brown, slightly shorter than length of hypopharyngeal sclerite bridge, broadly connected to one another to form a pale W-shaped or quadrate sclerite; epipharyngeal sclerite small, faintly pigmented, amorphous; anterior sclerite (Exley 1955) present in mature larvae; pharyngeal filter present, with 7 lamellate ridges extending the length of the pharynx.

Anterior spiracle (Figs. 5, 10) pale golden brown, projecting, usually with 8–10 tubules (but one had 7 and one had 11 on one side only; one had 11 on both sides) closely spaced in a single straight row; distal width 0.145-0.166 mm (n = 4), base cylindrical, about half as wide as distal width; tubules about as long as wide, rounded apically, each with a slitlike opening; felt chamber as in Fig. 10; unpigmented ecdysial scar posterior to tubules.

Segments T1–T3 and usually A1 with broken rows of weak, conical spinules on dorsal anterior margin, with 3–5, 3–5, 1–4, and 1–2 rows of spinules, respectively, at

dorsal midline; on T1 and T2 the spinulose area encircles the body, while on T3 the ventral spinulose area is separated from that of the dorsum; dorsal spinules absent on A2-A8; ventral spinulose areas on T1 with 10-12 rows, T2 and T3 with 3-7 rows each, and A1 with 4–7 rows of posteriorly directed spinules; ventrally, segments A2-A7 with 9-11 rows and A8 with 6-9 rows of spinules that are alternately arranged in groups of anteriorly and posteriorly directed rows, typically arranged as follows: 1-3 rows of small (approximately 0.005 mm long), anteriorly directed spinules that appear to overlap segmental lines and actually pertain to the preceding segment; 2-4 rows of small, posteriorly directed spinules; 1-2 rows of anteriorly directed spinules, and 2-5 rows of posteriorly directed spinules, some of which may reach 0.014 mm in length.

Thoracic segments with *Keilin's organ* (pedichaete) present as a trifid sensillum; other body sensilla exceedingly minute.

*Caudal segment* (Fig. 3) with a caudal ridge on the intermediate region, without a dark transverse line in the medial region; 10 pairs of sensilla present as follows: dorsal area with D1 and D2 on separate papillae very close to one another; lateral area with I3 and L on separate papillae; intermediate area with intermediate tubercle well developed, bearing the following sen-



Fig. 9. *Ceratitis rosa*, third instar, cephalopharyngeal skeleton (as, anterior sclerite; at, apical tooth; de, dorsal cornu; db, dorsal bridge; ds, dental sclerite; hs, hypopharyngeal sclerite; ls, fabial sclerite; md, mandible; pab, parastomal bar; phb, pharyngeal bar; st, subapical tooth; ts, tentoropharyngeal sclerite; vc, ventral cornu).

silla: 11a and 11b (on the same papilla or very close together, near medial end of caudal ridge), and 12 (below caudal ridge); ventral area with 3 V sensilla (one as a papilla sensillum and two as pit sensilla).

*Posterior spiracle* (Figs. 6, 11) above midline, with 3 slit-like openings, dorsal and central slit subparallel, ventral slit more medial and at an angle to the other two; rimae about 2.75–3.8 times longer than wide (0.065–0.082 mm long; 0.021–0.025 mm wide), separated from midline by approximately 2–3 times the length of the rima; spiracular processes well-developed, about half as long as rimae, mostly unbranched, some with 1–2 branches; numbers of trunks and tips as follows: 1 (dorsal) (8–9, 10–12), 11 (3, 5–6), III (4–7, 7–11), IV (ventral) (9–10, 11–14).

Anal lobes (Fig. 4) well-developed, protruding, entire (rarely grooved), surrounded by 2–4 broken rows of spinules.

Specimens examined.—SOUTH AFRI-CA: ex culture, Brian Barnes (n = 40, USNM, TAMU).

#### DISCUSSION

The larva of *Ceratitis rosa* shares the following characteristics with other Dacini for which larvae have been described: 1) a well-developed caudal ridge on the intermediate region of the caudal segment, 2) a stomal organ borne on a small primary lobe that is surrounded by secondary stomal lobes, 3) no sclerotized stomal guards below the stomal organ, and 4) dental sclerites present posterior to the base of the mandible.

Like other Ceratitidina, it has: 1) the area lateral to the oral ridges without accessory plates and 2) the secondary stomal lobes broad, flat, and leaf-like (as also in *Bactrocera (Bactrocera)*), rather than having the medial secondary stomal lobes similar to oral ridges (as in *Dacus*, *B. (Zeugodacus)* and related subgenera). Aside from the dental sclerites (which require slide-mounting), all these structures are sufficiently discernible in mature larvae in good condition, with a dissecting microscope at a magnification of approximately  $750\times$ , to allow identification to the generic level.

Numbers of oral ridges and anterior spiracular tubules, in addition to the caudal ridge (described as a row of papillae by Phillips (1946) and Berg (1979)), have been among the characters most commonly used for the identification of larvae of *C. capi*-



Figs. 10–13. Third instar spiracles of *Ceratitis rosa* and *C. capitata*. 10–11, *C. rosa*. 12–13, *C. capitata*. 10, 12, Anterior spiracles, 11, 13 Posterior spiracles (ri, rima; sp. spiracular processes).

*tata.* However these characters are not useful in distinguishing the larva of *C. capitata* from that of *C. rosa*, due to virtually complete overlap in numbers of oral ridges

(usually 9–11, rarely 8 or 12 in both species) and anterior spiracular tubules (usually 8–10, rarely 7 or 11 in both species). Extremes of oral ridge count data in the sample of *C. capitata* included one specimen with 8 ridges and six specimens with 12 ridges on one side only, and one specimen with 12 ridges on both sides. Extremes of anterior spiracular tubule count data for *C. capitata* included a single specimen with 6 tubules on one side (spiracle plainly deformed), three with 7 tubules and two with 11 tubules on one side only.

The larva of C. rosa is distinguishable from that of C. capitata by having the area of the cephalic lobes around the maxillary palps more round and proluberant, rather than slightly flattened in lateral view (Fig. 7), anterior spiracles basally narrower, with sides subparallel (Fig. 10), rather than flaring from the base (Fig. 12), and posterior spiracular slits (Fig. 11) generally less robust than in C. capitata (Fig. 13). It also has a minute subapical tooth on the mandible (Fig. 9), which is clearly visible by SEM and in slide-mounted material, which is never present in C. capitata. Ceratitis rosa lacks spinules on the dorsal anterior margin of segment A2, while C. capitata frequently has one or two isolated lines of spinules on this segment.

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